

q•Hot Taq DNA Polymerase Kit with 10X Buffer B1 or B2

for research and in vitro use only

Protocol

1. Thaw and then centrifuge all kit components
2. Prepare your reactions using the Recommended Reaction Mix
3. Centrifuge your samples
4. Perform PCR using the Recommended Cycling Settings

Recommended Reaction Mix			
COMPONENT	CONC.	AMOUNT	FINAL CONC.
q•Hot Taq ¹	5 U/μL	0.2 - 0.5 μL	1 - 2.5 U
Buffer B1 or B2	10X	5 μL	1X
MgCl ₂	25 mM	3 - 5 μL	1.5 - 2.5 mM
dNTP mix	20 mM	0.5 μL	0.2 mM
Primer, F	10 pmol/μL	0.5 - 1.5 μL	100 - 300 nM
Primer, R	10 pmol/μL	0.5 - 1.5 μL	100 - 300 nM
Template DNA		5 - 500 ng	0.1 - 10 ng/μL
Enhancer	10X	0 - 15 μL	0 - 3X
Nuclease-free H ₂ O		variable	
Total		50 μL	

¹ q•Hot Taq is suitable for amplicons up to 3kb. For amplification of larger DNA fragments, use qARTA•Hot Master Mixes.

Recommended Cycling Settings			
CYCLE STEP	TEMP (°C)	TIME	CYCLES
Initial denaturation	95	12 - 15 min	1
Denaturation	95	30 - 60 s	26 - 35
Annealing ¹	T _M - 4	30 - 60 s	
Extension ²	72	1 min/kb	
Final extension	72	5 - 10 min	1

¹ Set annealing temperature to be 4°C lower than T_M of primers or set it to 50-68°C initially.
² Use amplicon length to optimize extension time: approx. 1 min per 1000 bases.

Estimating primer melting temperature:
 For primers containing less than 25 nucleotides, T_M = 4 (G + C) + 2 (A + T), where G, C, A, T represent the number of respective nucleotides in the primer. If primers contain more than 25 nucleotides use specialized software to calculate T_M.

q•Hot Taq Order Information

Cat. No.	Size	Conc.	No. Rxns 50 μL each	q•Hot Taq	10X Buffer B1 ¹ (Mg ²⁺ & detergent free)	10X Buffer B2 ² (Mg ²⁺ free)	25 mM MgCl ₂	10X Enhancer	20mM dNTP Mix
QH-S	100 U	5 U/μL	40	20 μL	0.5 mL	0.5 mL	0.5 mL	0.1 mL	20 μL
QHB1-500	500 U	5 U/μL	200	100 μL	2.4 mL	N/A	2.4 mL	0.1 mL	100 μL
QHB2-500					N/A	2.4 mL			
QHB1-1000	1000 U	5 U/μL	400	200 μL	4.8 mL	N/A	4.8 mL	0.5 mL	200 μL
QHB2-1000					N/A	4.8 mL			

¹ Buffer B1 is a 10X Reaction Buffer, Mg²⁺ and detergent free: Tris-HCl, (NH₄)₂SO₄

² Buffer B2 is a 10X Reaction Buffer, Mg²⁺ free: Tris-HCl, (NH₄)₂SO₄, and detergent

Some applications of this product are covered by patents issued to parties other than qARTA Bio, and may require a license which is not provided by the purchase of this product. User should obtain a patent license if appropriate.

Description

q•Hot Taq is a chemically modified DNA polymerase of the thermophilic bacterium *Thermus aquaticus*. The enzyme is activated by 15 minutes of incubation to prevent non-specific annealing. It catalyzes 5'→3' synthesis of DNA and has no detectable 3'→5' exonuclease activity.

Definition of activity unit

One unit is defined as the amount of enzyme required to catalyze the incorporation of 10 nmol of deoxyribonucleotides into a polynucleotide fraction in 30 minutes at 74°C.

Storage and dilution buffer

50% glycerol (v/v), 20 mM Tris-HCl pH 8.7 at 25°C, 100 mM KCl, 0.1 mM EDTA and stabilizers.

Quality control

The enzyme is free of nicking and priming activities, exonucleases and non-specific endonucleases. Activity and stability tested via functional assay. The error rate per nucleotide per cycle is ~2.5 x 10⁻⁵ and accuracy is ~4 x 10⁴.

Eco-friendly shipping and storage

- Shipped at ambient temperature without ice
- Store at -20°C upon arrival